

pTNMAX (general vector)

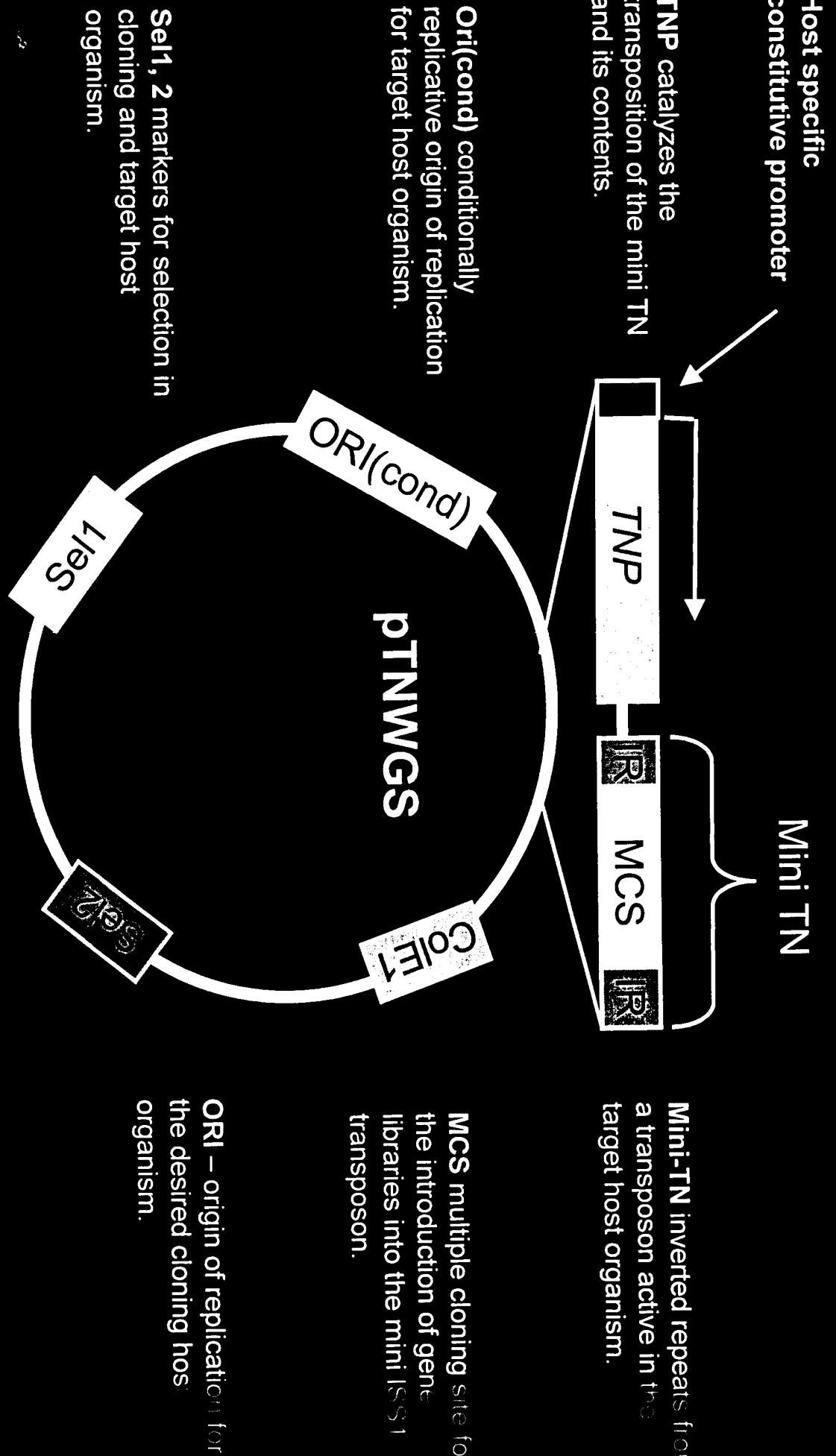
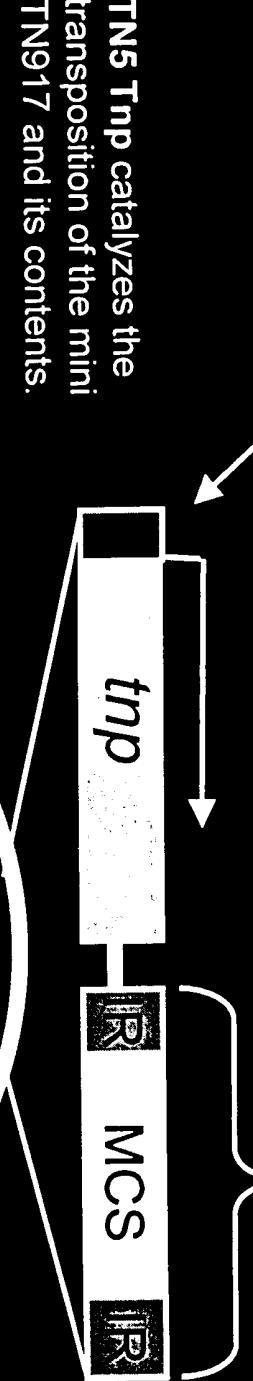


Figure 1A

pWGS:5

Host specific
constitutive promoter

Mini TN5



Mini TN5 Tn5 inverted repeats flanking a multiple cloning site into which gene libraries can be cloned.

Ori-ts temperature sensitive origin of replication for plasmid maintenance in target bacteria (gram positive or negative bacteria).

Kan^r confers resistance to kanamycin to *E. coli*

Erm^r confers resistance to erythromycin in Gram positive bacteria.

Figure 1B

pWGS:917

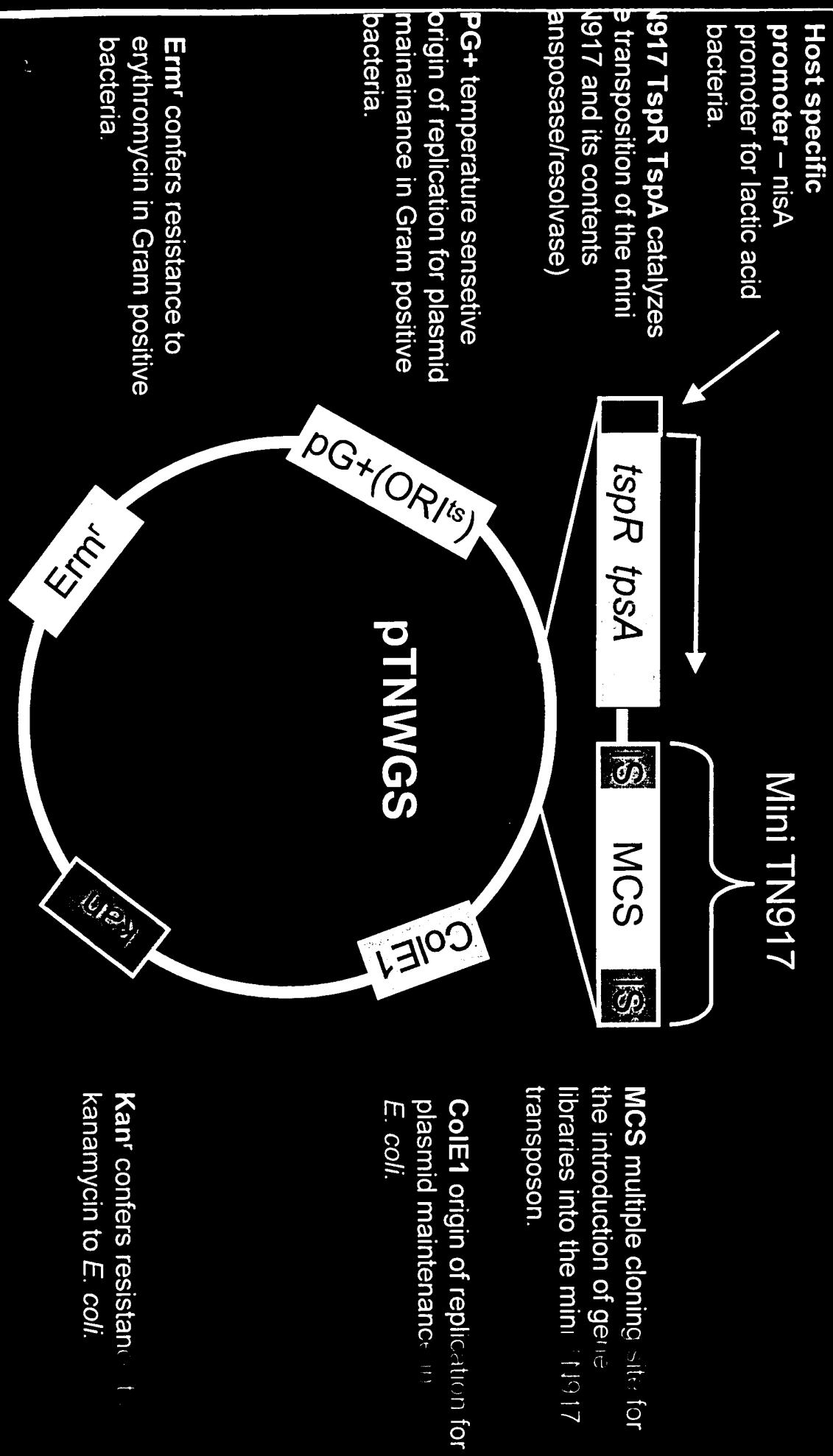
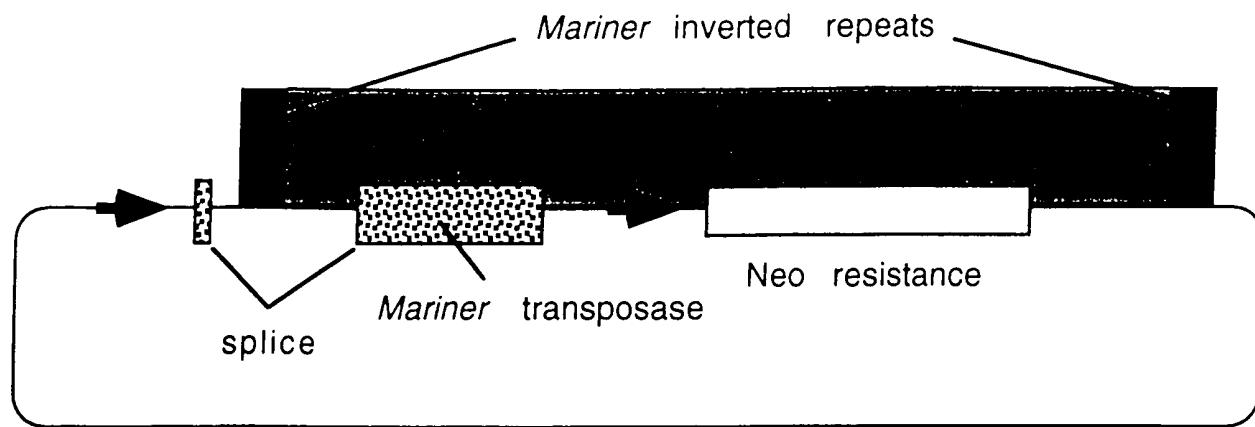


Figure 1C

Figure 2

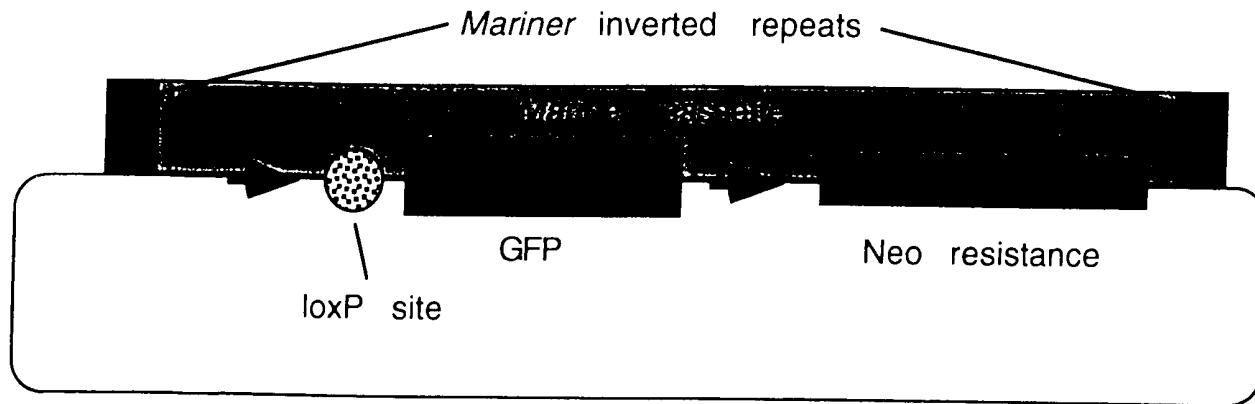
A

Efficient integration into mammalian cells using evolved *Mariner* transposons



B

Mariner transposon for inserting loxP sites at loci with desirable expression properties



Methodology for Isolating Hosts with improved Phenotypes by Whole Genome Shuffling (WG_S)

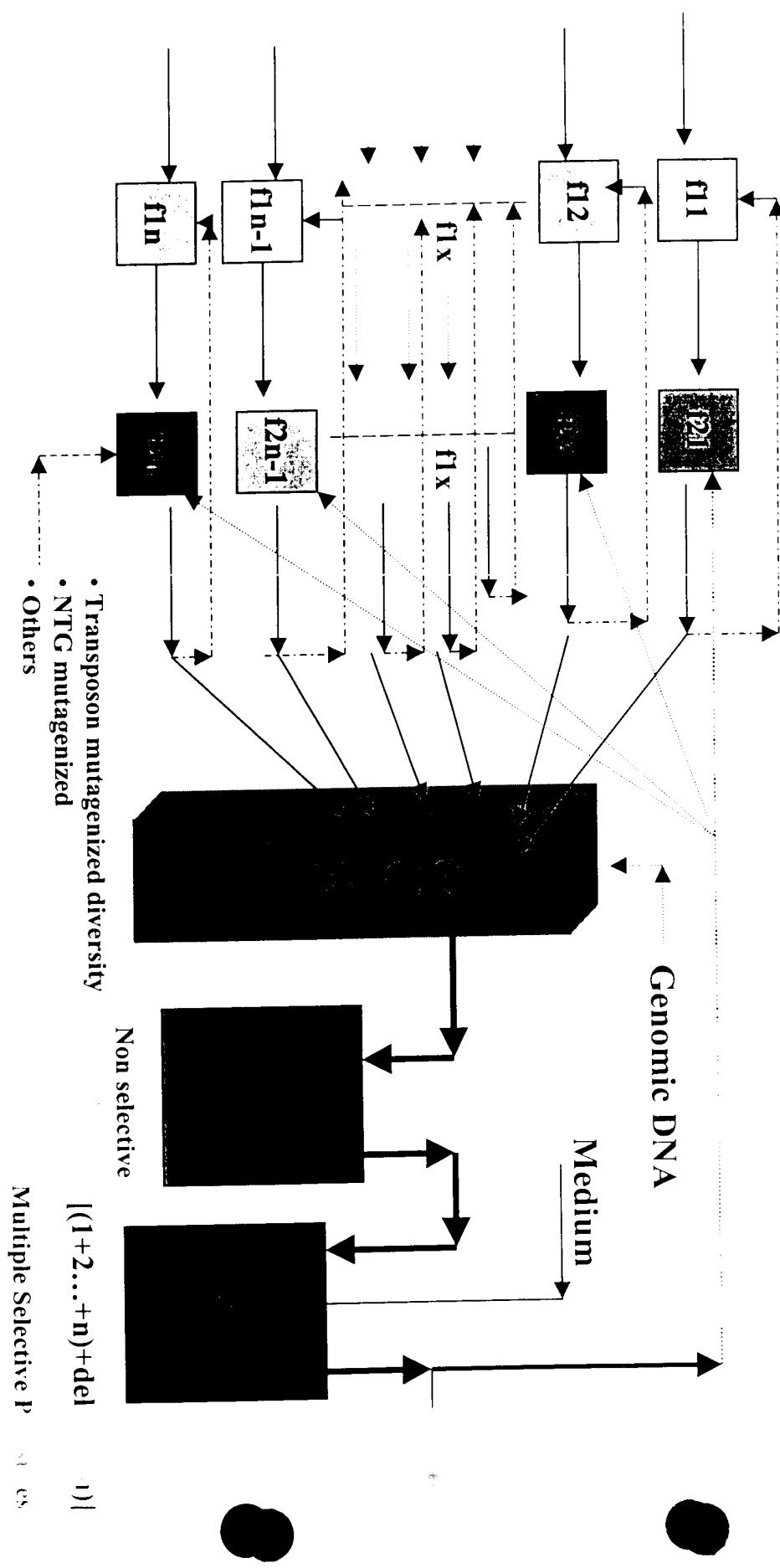
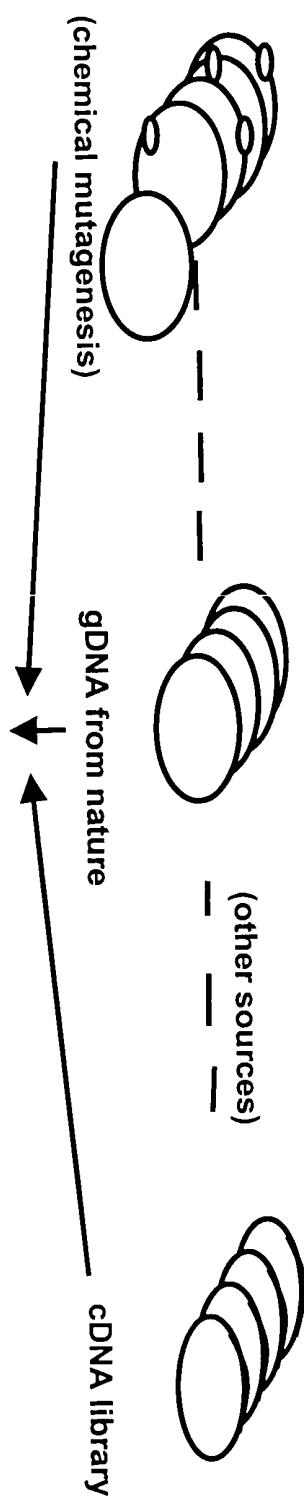


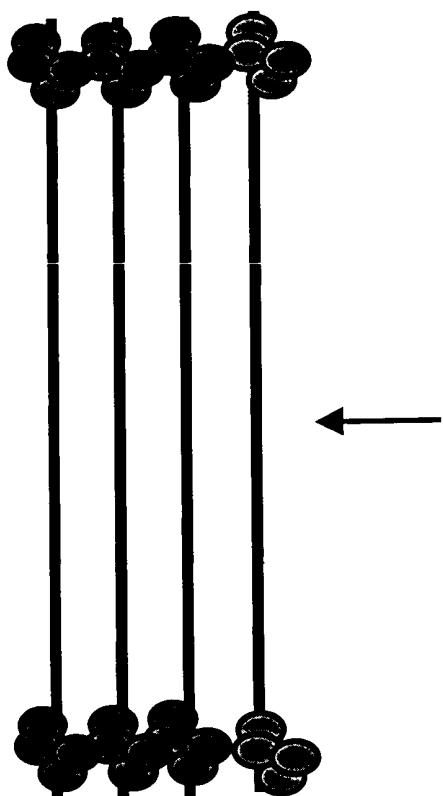
Figure 3

Shuffling of Genomes *In Vitro*: Formation of transposomes

sources of genomic diversity



1. generate subgenomic fragments
2. clone within Tn5 transposon ends
3. add Tn5 transposase



* “transposomes”: complexes poised for integration upon exposure to Mg⁺⁺.

Figure 4A

Shuffling of Genomes *In Vitro*:
Breeding multiple donor genomes with a single acceptor genome

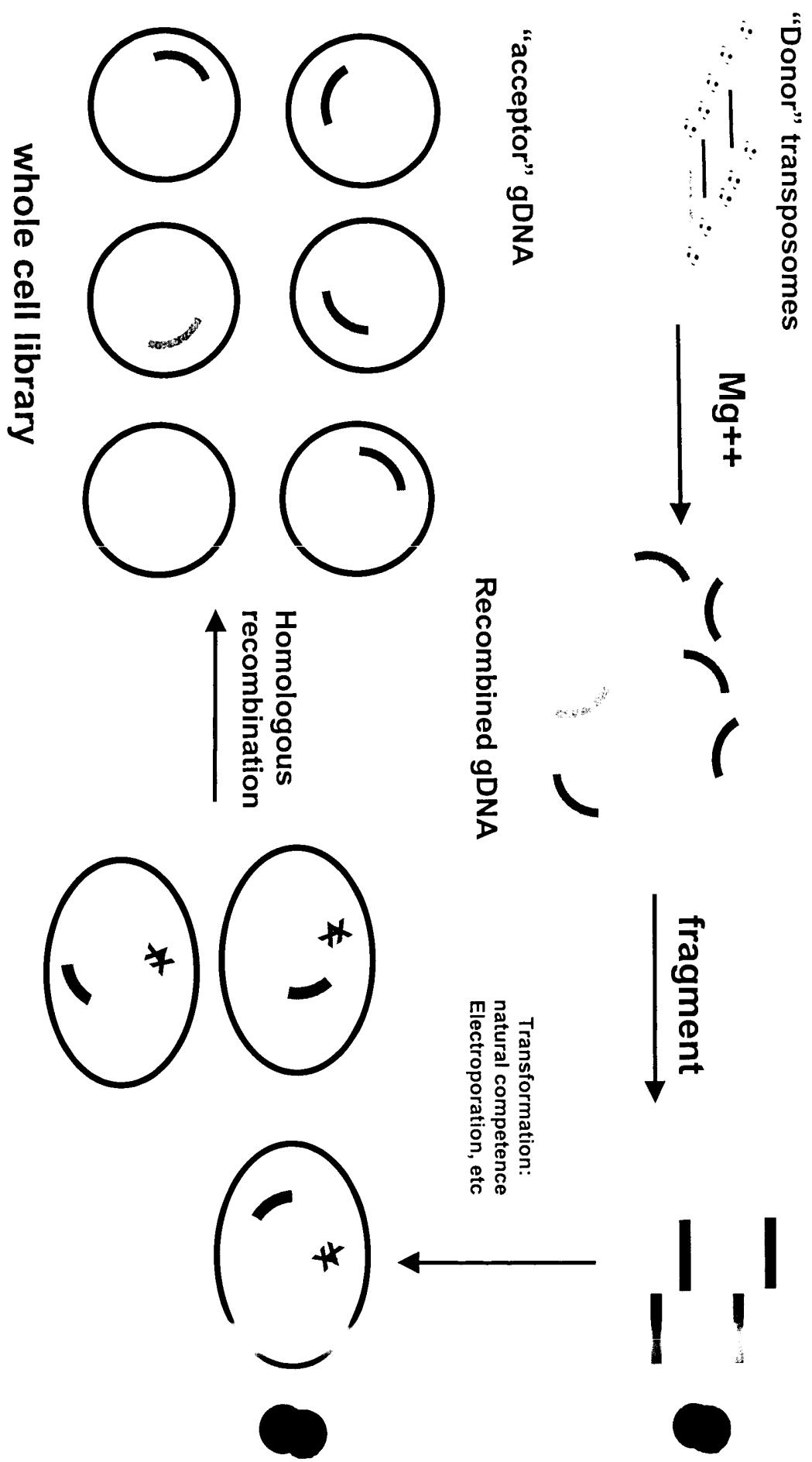


Figure 4B

Shuffling of Genomes *In Vitro*:
Breeding multiple donor genomes with multiple acceptor genomes

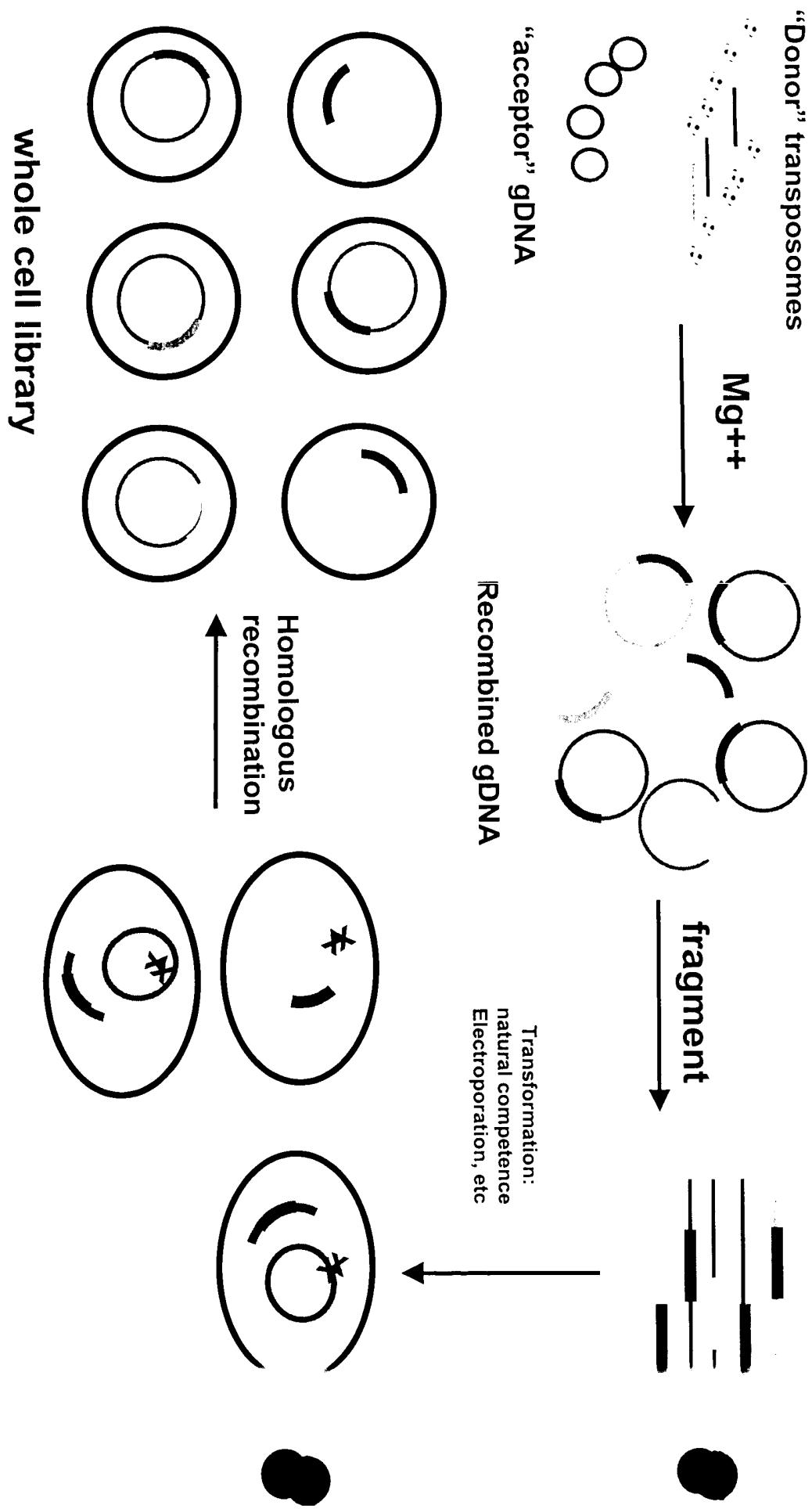


Figure 4C

Shuffling of Genomes *In Vitro*:
Split pool recursive *in vitro* recombination of multiple genomes

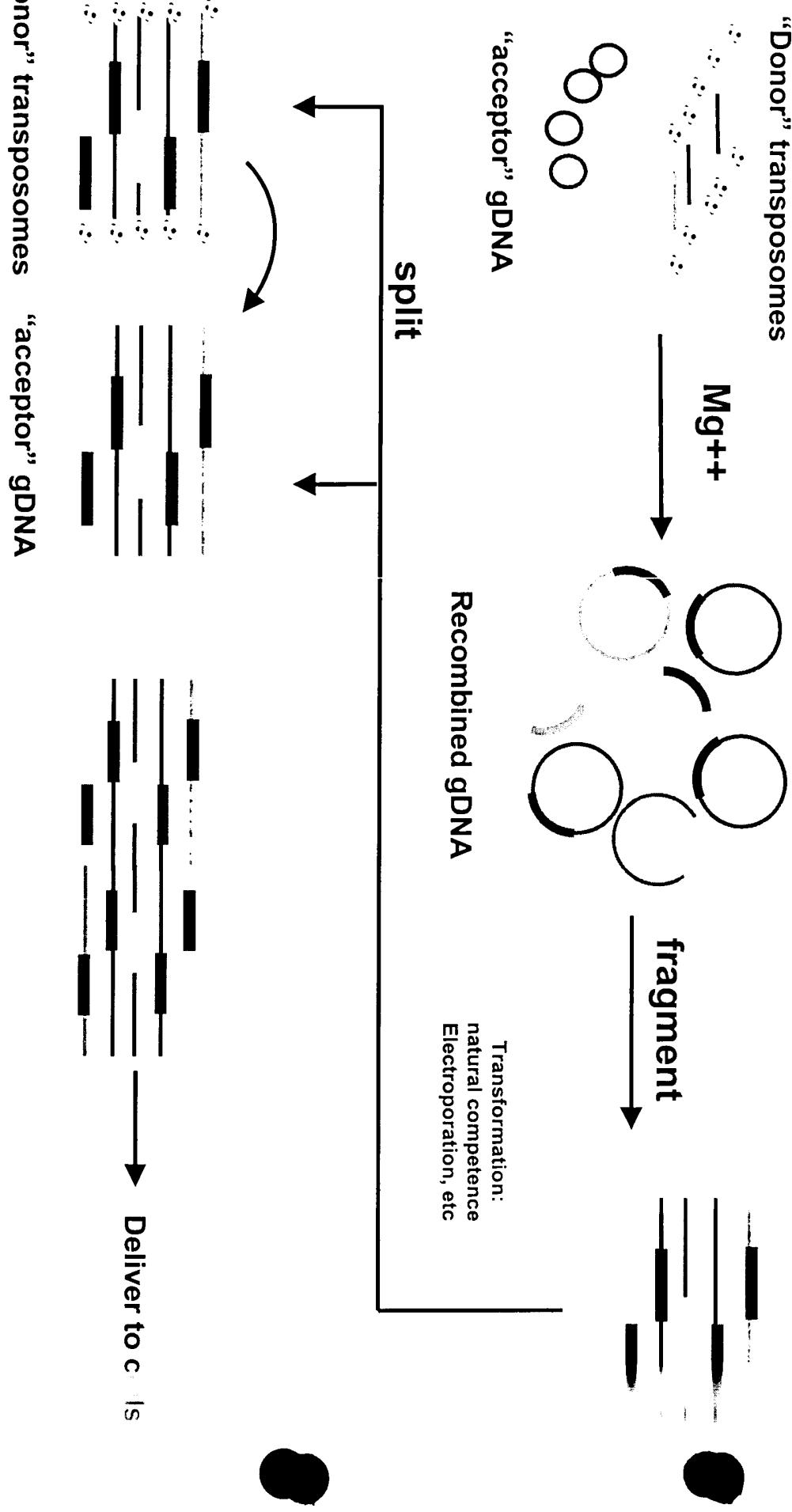


Figure 4D